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The Potential of Natural Deep Eutectic Solvents for the Extraction of Phenolic Compounds and Caffeine from Green Coffee (*Coffea arabica*) Beans

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ABSTRACT

A natural deep eutectic solvent (NDES) mixture was studied for extracting phenolic compounds from green coffee beans (GCB) (*Coffea arabica*). Dehydrated, finely ground GCBs were extracted with water, 50 % aqueous ethanol, and NDES (glucose and glycerin, 1:1 M) at 30 ˚C for 30, 60, and 90 min using a shaking incubation method. The process was repeated at 50 °C for 30 min. Total phenolic content (TPC) was measured via the Folin-Ciocalteu test. Chlorogenic acids (3-, 4-, and 5-caffeoylquinic acid) and caffeine were quantified by HPLC-PDA. At 30 °C, the highest TPC (48.7 ± 0.6 mg GAE/g) was found in the NDES 30-min extract, comparable ($P > 0.05$) to NDES 60-min (47.4 ± 0.1 mg GAE/g) and 90-min (48.6 ± 1.1 mg GAE/g) extracts. The highest chlorogenic acid content (52.4 ± 0.6 mg/g) was in 50 % ethanol, 30-min extraction, similar ($P > 0.05$) to other ethanolic extracts and NDES 60- and 90-min extracts. Caffeine content (12.1 ± 0.2 mg/g) peaked in 50 % ethanol, 30-min extract, similar ($P > 0.05$) to most other extracts except NDES 30-min (11.3 ± 0.3 mg/g). NDES at 50 °C gave 46.9 ± 0.5 mg/g TPC, 48.6 ± 0.7 mg/g chlorogenic acid, and 11.1 ± 0.2 mg/g caffeine, not significantly different from 30 °C. These results indicate glucose–glycerin NDES is a promising, eco-friendly solvent for extracting phenolic compounds from GCBs, providing an alternative to 50 % ethanol.
Keywords: chlorogenic acid, deep eutectic solvents, green coffee beans, phenolic compounds

1. INTRODUCTION

Green coffee bean (GCB) has received much attention in recent studies for its potential health benefits in weight management, heart health, neurological conditions, and blood sugar regulation, due to its profile rich in several bioactive compounds, such as caffeine, diterpenes, chlorogenic acids, ferulic acid, caffeic acid, p-coumaric acid, and trigonelline. Therefore, GCB extracts are widely

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used as supplements to treat or prevent several non-communicable diseases, such as type 2 diabetes, high blood pressure, and cardiovascular disease (Munyendo et al., 2021; Khalili-Moghadam et al., 2023). Caffeine, which is an alkaloid, is responsible for 1-2.5 % of coffee dry matter composition. Additionally, caffeoylquinic acids (CQA), such as *3*-CQA, *4*-CQA, and *5*-CQA, that belong to the group of chlorogenic acids, are responsible for 4-12 % of the dry matter coffee composition (Farah et al., 2005). Amongst these phytochemicals, *5*-CQA is the main phenolic compound with redox properties that accumulates in coffee beans during the maturation process (Acidri et al., 2020; Tripathi et al., 2024). Previous animal-based and human clinical studies have shown that GCB extracts exhibit anti-obesity and anti-diabetic properties, attributed to the lipid-lowering and insulin-sensitivity-improving effects of *5*-CQA (Sudeep & Prasad, 2021; Khalili-Moghadam et al., 2023; Chandimala & Ajtony, 2025). However, as coffee is typically consumed after roasting, a considerable amount of these phytochemicals is degraded. For example, Acidri et al. (2020) reported that roasting GCBs resulted in a 66 % decline of their initial phytochemical content. However, the negative impact of roasting on caffeine and trigonelline was lower than that of *5*-CQA. Therefore, when developing extracts to generate health benefits, GCB extracts are more imperative and beneficial than roasted coffee extracts.

Solvent extraction is the most studied method for isolating phytochemicals with antioxidant potential from plant sources. A range of factors, such as temperature, time, and solvent concentration and polarity, influence the extraction and purification of bioactive compounds from plant materials. The type and concentration of bioactive compounds in the extract mainly depend on the chemical composition of the solvent system. Overall, solvents with medium polarity, such as ethanol, methanol, and acetone, have been shown to give higher yields of total phenolics and antioxidant activity in GCB extracts (Tripathi et al., 2024).

Recent developments in green chemistry have encouraged the creation of new organic solvent systems, such as natural deep eutectic solvents (NDES), as safer alternatives to harmful organic solvents. NDES are made by mixing hydrogen bond donors (e.g., organic acids, sugars) with hydrogen bond acceptors (e.g., choline chloride). The NDES mixture usually has a freezing point lower than that of its components (Loukri et al., 2022). NDES are considered environmentally friendly solvents because they are derived from non-toxic and biodegradable natural ingredients like sugars, salts, amino acids, and organic acids. Their ability to dissolve a wide range of biomolecules with excellent solubility and thermal stability, coupled with low toxicity and cost, makes them a promising green solvent for extracting phytochemicals from various plant materials (Ruesgas-Ramón et al., 2017; Okeke et al., 2025). However, research on the extraction of GCB phenolic compounds using NDES and their food applications remains limited in the current literature. For example, aqueous ethanolic extracts of GCB are the most commonly used for food applications, such as enhancing the microbial and oxidative stability of meat products (Atondo-Echeagaray et al., 2025). Therefore, this study focused on preparing GCB extracts using an NDES mixture with a shaking incubation extraction method and on comparing the phenolic content, *5*-CQA, *3*-CQA, *4*-CQA, and caffeine levels with ethanolic and water extracts. Based on previous studies involving NDES in plant extracts (Rumiyati et al., 2024; Sik et al., 2024), a mixture of glucose (hydrogen bond donor) and glycerin (hydrogen bond acceptor) was selected as the NDES system for this research.

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2. MATERIALS AND METHODS

2.1 Materials and equipment

Commercial GCB (*Coffea arabica*) samples used for the measurements were purchased from Gourmet Kave, Hungary. The Folin-Ciocalteu reagent and gallic acid were purchased from Merck (Germany). Anhydrous sodium carbonate and d-(+)-glucose anhydrate were purchased from Riel-de Haan (Germany). Glycerol was purchased from Sigma Aldrich, Hungary. Moreover, absolute ethanol was purchased from VWR chemicals (France). The standard *5*-CQA and caffeine (purity ≥ 95 %) were purchased from Merck Science Life Ltd. (Budapest, Hungary). For the HPLC analysis, acetonitrile of HPLC grade (Fisher Scientific, UK) and ortho-phosphoric acid of analytical grade (Lachner, Hungary) were used.

A 70 M model food dehydrator (Precision Scientific, United States) was used for drying GCBs. Dried coffee beans were ground into fine particles with a coffee grinder (Aroma, Hungary). The extraction of phenolic compounds from GCBs was carried out using a shaking incubator (GFL 3032, Germany). The samples were centrifuged using a Z206A laboratory centrifuge (Hermle, Germany). For the experiments, deionized ultra-pure water was obtained from an ultra-pure water system (Milli Q-SQ 2 series, Germany). Spectrophotometric analysis was implemented using a Spectroquant Pharo 100 spectrophotometer (Merck, Germany). The instrumental analysis of the *5*-CQA and caffeine was performed using an HPLC-PDA system (Jasco, Japan) equipped with a 4-line degasser (Jasco DG-2080-54), intelligent HPLC pump (Jasco PU-980), ternary gradient unit (Jasco LG-980-02), intelligent sampler (Jasco AS-2055), column thermostat (Phenomenex TS-130), and column compartment. ChromeNAV software (Jasco, Japan) was used for data acquisition.

2.2 Methodology

2.2.1 NDES preparation

The NDES mixture was prepared according to Sik et al. (2024). Glucose and glycerin were mixed at a 1:1 M ratio with 10 mL of water and stirred at 500 rpm (approximately 3 h) at a temperature of 90 °C until a viscous, colorless liquid was obtained. The resulting solvent was mixed with 50 % (w/w) high-purity deionized water, cooled down to room temperature, and used for the extraction.

2.2.2 Extraction conditions

A 0.25 g powdered GCB sample was mixed with 5 mL of glucose-glycerin-based NDES. Then, samples were placed in a shaking incubator set to 30 °C at 150 rpm for varying times: 30, 60, and 90 min. Additionally, the effect of using a higher temperature-short time extraction was tested by applying a 50 °C temperature at 150 rpm for 30 min, following the same procedure. After extraction, the samples were diluted fivefold with deionized water and centrifuged at 6000 rpm for 10 min, and the filtered supernatants were used for the spectrophotometric/HPLC analysis. Solvent volume and dilution were selected after pre-trials. To facilitate comparisons, water extracts and 50 % (v/v) ethanolic extracts were prepared under the same extraction conditions.

2.2.3 Determination of total phenolic content (TPC)

The TPC of the extracts was measured by the Folin-Ciocalteu method as described by Sik et al. (2024) with slight modifications. A 100 µL sample was mixed with 1.5 mL of deionized water, 10 % (v/v) Folin-Ciocalteu reagent, and 2 mL of sodium carbonate solution (7.5 g/100 mL), respectively. After

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90 min of incubation at room temperature, the absorbance was measured at 725 nm versus the blank. A standard curve was developed by plotting gallic acid concentration (ranging from 0-1000 µg/mL) versus absorbance based on spectrophotometric analysis. The regression equation for TPC was y = 1.3005x + 0.0152, with a high correlation coefficient (0.999). The standard curve met the standards, with a 0.0261 standard error of slope, a 0.056 mg/mL LOD, and a 0.171 mg/mL LOQ, and was used to calculate TPC yields for each extract. TPCs were expressed as mg of gallic acid equivalents per gram of dry plant material (mg GAE/g).

2.2.4 Determination of chlorogenic acid and caffeine contents

Working standard solutions of 5-CQA and caffeine with concentrations ranging from 0 to 1000 µg/mL and 0 to 500 µg/mL, respectively, were prepared and injected into the HPLC system under specified conditions to develop calibration curves for 5-CQA and caffeine by plotting concentration against peak area. Separation was achieved using an InertSustain Phenyl column with 5 µm pore size, 3 mm diameter, and 150 mm length, maintained at 35 ˚C. The mobile phase consisted of 7 % acetonitrile and 93 % water containing 0.1 v/v% phosphoric acid. The flow rate and injection volume were set at 0.8 mL/min and 5 µL, respectively. Identification of 5-CQA and caffeine was performed by comparing their retention times with those of the respective standards and by spiking samples with small amounts of the appropriate standards. Quantification in each extract was conducted using the regression equation of the best-fit line from the calibration curves. Due to the unavailability of 3-CQA and 4-CQA standards in our lab, their quantification was based on the area of the 5-CQA standard combined with the molar extinction coefficients of the respective CQA, as previously described by Farah et al. (2005).

2.2.5 Research design and statistical analysis

All experiments were conducted using a one-factor-at-a-time (OFAT) approach, with three replicates per factor. All data were presented as the mean ± standard deviation of triplicate analyses. Minitab®19 statistical software was used for the analysis of variance (ANOVA) at a 95 % confidence level to determine the significance of differences. All tested parameters followed normal distribution based on designed probability plots ($P > 0.05$, Anderson-Darling value < 0.75), and Tukey's HSD test was used to compare all possible pairs of means to determine whether the differences were statistically significant.

3. RESULTS

3.1 Total phenolic content in GCB extracts

TPC values detected in the three solvent systems – water, 50 % ethanol, and a glucose-glycerin-based NDES mixture – are shown in *Figure 1*. The highest TPC yield (48.7 ± 0.6 mg GAE/g) was observed in the NDES extract with a 30-min extraction time, followed by the NDES extract with a 90-min (48.6 ± 0.6 mg GAE/g) extraction. However, the TPC yield was similar ($P > 0.05$) in all tested extraction times for the NDES. According to the one-way ANOVA test, NDES yielded significantly higher levels than 50 % ethanol (45.8 ± 1.1 mg GAE/g) at a 30-min extraction time ($P < 0.05$). Similarly, NDES yielded significantly higher amounts than water (41.9 ± 1.1 mg GAE/g) at 30 min ($P < 0.05$). These results suggest that glucose-glycerin-based NDES can effectively replace water and organic solvents, such as ethanol, for the extraction of phenolic compounds from GCBs.

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Overall, the TPC content observed in all solvent extracts in this study (4.0-4.8 %) exceeds the previously reported TPC yield in water extracts (3.8 %) of GCBs (Priftis et al., 2015).

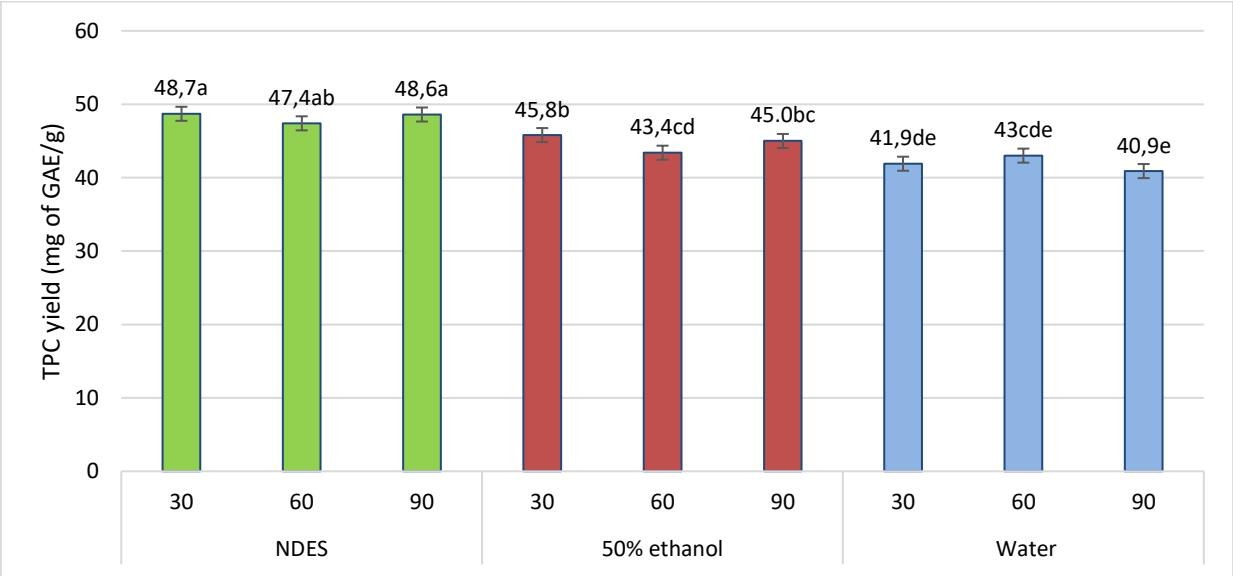

Figure 1: Total phenolic content of GCB extracts at 30 °C temperature

Values denote the mean of the three replicates. Means followed by the same letter are not significantly different at a 95 % confidence level according to Tukey's HSD test. Letters are assigned from a to e in descending order of the mean values.

3.2 Chlorogenic acid concentration in GCB extracts

Figure 2 depicts the representative base-peak chromatogram of the NDES extract for 30 min, analyzed with the InertSustain Phenyl HPLC column. When the phenyl column was used for the analysis at the occupied wavelength of 325 nm, peak 2 was identified as *5*-CQA by comparing it with the standard *5*-CQA spectra. Based on the literature on peak shape and peak purity when analyzing UV spectra, peaks 1 and 3 were identified as *3*-CQA and *4*-CQA, respectively. All three peaks had a peak purity level of above 99.9 %, confirming that peaks 1 and 3 are isomers of *5*-CQA. Peak 4 also showed a purity above 90 %, although it was not identified in this study due to a lack of a standard and information in the literature.

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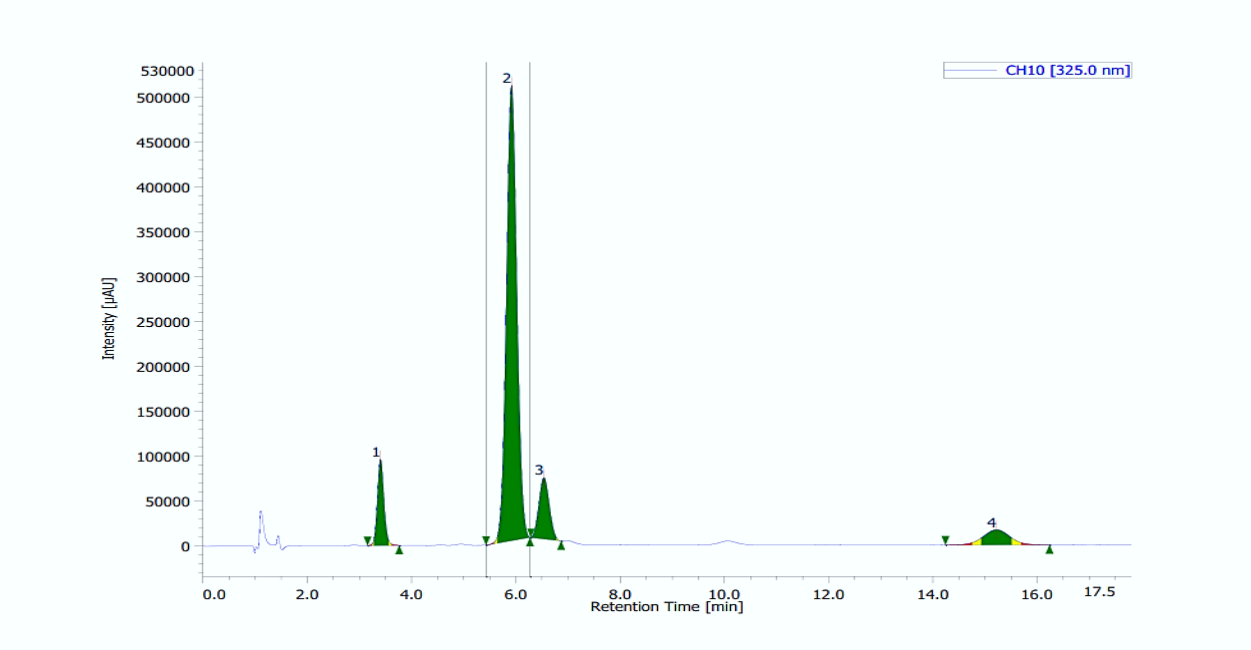

Figure 2: Elution order of CQA isomers in an NDES extract (30 °C for 30 min) of GCBs with InertSustain phenyl column at detector wavelength of 325 nm

Peaks 1, 2, 3, and 4 represent 3-CQA, 5-CQA, 4-CQA, and unidentified, respectively. The mobile phase contained 7 % acetonitrile and 0.1 % phosphoric acid. Flow rate was 0.8 mL/min. The purity of 1-3 peaks was > 99.9 %. The green color represents high purity at the peak's top position.

Concentrations of 5-CQA, 3-CQA, and 4-CQA in GCB extracts at 30 °C temperature are represented in *Figure 3*. The highest 5-CQA concentration (41.5 ± 0.5 mg/g) was observed in ethanolic extraction for 30 min. However, there was no significant difference ($P > 0.05$) in the 5-CQA content among all ethanolic extracts and NDES extracts at 60 and 90 min. NDES extraction for 30 min indicated 38.9 ± 0.9 mg/g of 5-CQA yield, which was significantly lower ($P < 0.05$) than the ethanolic extraction for 30 min. Water extracts yielded the lowest 5-CQA levels (34.3-34.8 mg/g). A similar pattern was observed for 4-CQA as well. The highest 4-CQA yield (6.2 ± 0.1 mg/g) was observed with ethanolic extraction for 30 min, which was not significantly different from the NDES extraction yields at 60 and 90 min. NDES extraction for 30 min indicated 5.8 ± 0.2 mg/g of 4-CQA yield. Water extracts yielded the lowest 4-CQA content (5.2 mg/g). Regarding 3-CQA concentration, there was no significant difference ($P > 0.05$) between all ethanolic (5.1-5.2 mg/g) and NDES extracts (5.0 mg/g). However, in all water extracts (4.4 mg/g), the 3-CQA yield was significantly lower than the above.
Overall, the highest chlorogenic acid concentration was observed in ethanolic extraction for 30 min (52.4 ± 0.6 mg/g). This value was similar ($P > 0.05$) to other ethanolic extraction yields and NDES extraction for 60 and 90 min. NDES extraction for 30 min indicated 49.7 ± 1.2 mg/g of total chlorogenic acid yield, which was significantly lower ($P < 0.05$) than the ethanolic extraction for 30 min. Water extracts yielded the lowest total chlorogenic acid (43.9-44.4 mg/g). These values align with the documented range of 3 % to 6 % of CQAs in C. arabica dry matter (Farah & Donangelo, 2006). The present study reveals that 5-CQA is the predominant phenolic compound in NDES, ethanolic, and water extracts of GCBs.

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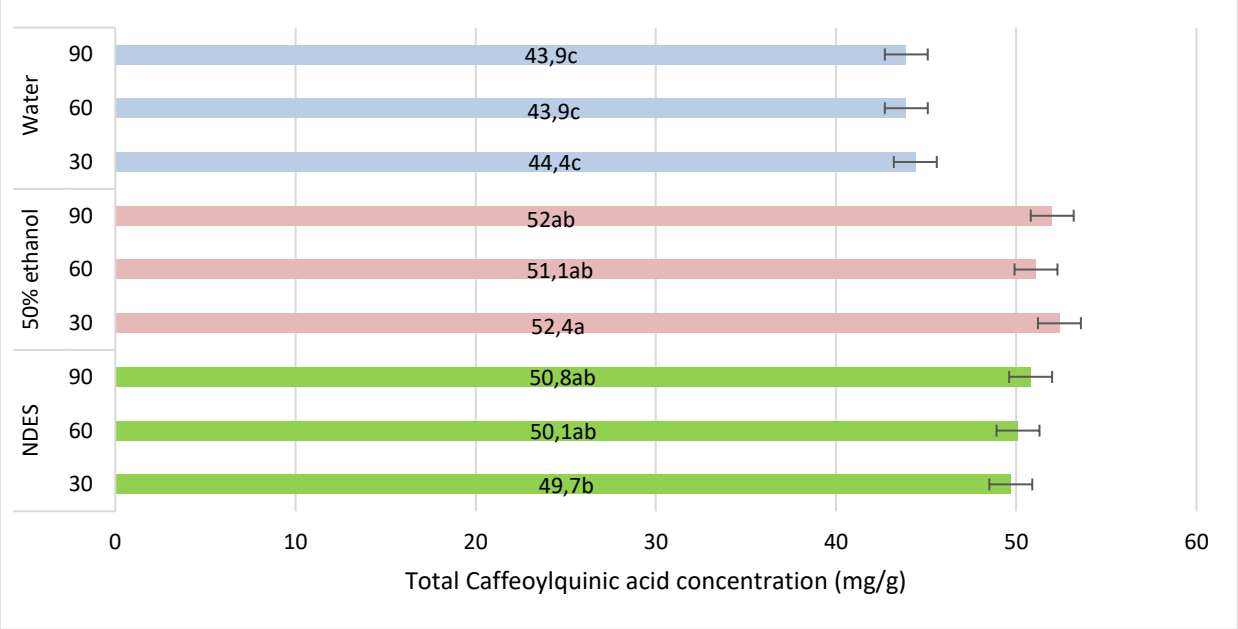

Figure 3: Total caffeoylquinic acid concentration in green coffee bean extracts

Values denote the mean of the three replicates. Means followed by the same letter are not significantly different at a 95 % confidence level according to Tukey's HSD test. Letters are assigned from a-c based on the descending order of their mean values, respectively.

3.3 Caffeine concentration in GCB extracts

There was no significant difference ($P > 0.05$) between the water extracts and NDES extracts for the caffeine yield. The highest caffeine content (12.1 ± 0.2 mg/g) was observed in 50 % ethanol 30-min extraction. Ethanolic extracts showed comparatively higher caffeine yields than other extracts. However, only the NDES extraction for 30 min showed a significantly lower yield than the 50 % ethanol extraction for 30 min (*Figure 4*).

In a study, Loukri et al. (2022) reported higher caffeine yields from coffee pulp using choline chloride and glycerin-based NDES solvent systems compared with the aqueous ethanolic system. In contrast, in the present study, ethanolic extraction for 30 min yielded significantly higher caffeine than all NDES extracts.

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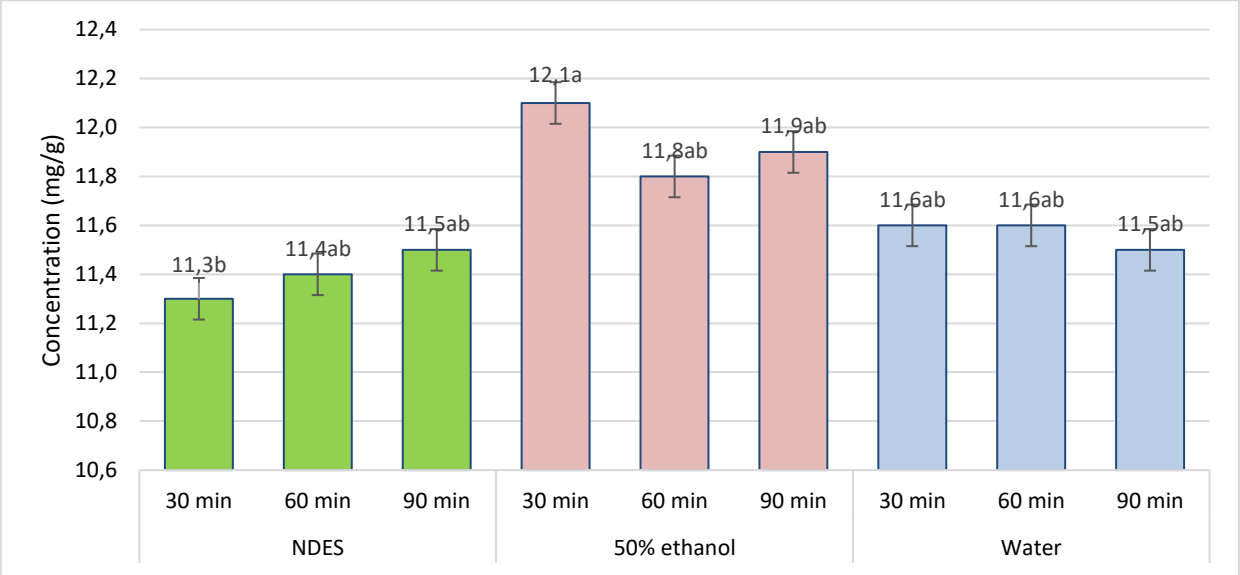

Figure 4: Caffeine content of GCB extracts at 30 °C temperature

Values denote the mean of the three replicates. Means followed by the same letter are not significantly different at a 95 % confidence level according to Tukey's HSD test.

3.4 Effect of temperature on the concentration of phenolic compounds and caffeine

The effect of using a higher temperature (50 °C) for a 30-min extraction was also examined in this study. The TPC, total CQA and caffeine yields compared to 30 °C are shown in *Figure 5*. According to the results, the TPC yield tends to decrease at 50 °C compared to 30 °C in NDES and ethanol extracts, although a slight increase was observed in water extracts, which was not significant ($P > 0.05$). The total CQA yield also slightly decreased in NDES extracts when the temperature was raised from 30 °C to 50 °C. However, water and ethanolic extracts showed an increase in total CQA yields with increasing temperature. This increase was not significant in ethanol, whereas it was significant in water extracts. Furthermore, *Figure 5* illustrates that the total CQA concentration exceeds the total phenolic concentration in all GCB extracts, regardless of the extraction temperature. The difference between TPC and CQA is more prominent in ethanolic extracts than in NDES and water extracts. Moreover, the figure underscores that the yield of caffeine, an alkaloid, is significantly lower than the yields of phenolics in all GCB extracts. Additionally, caffeine yield was lower at higher temperatures across all solvent types used in this study.

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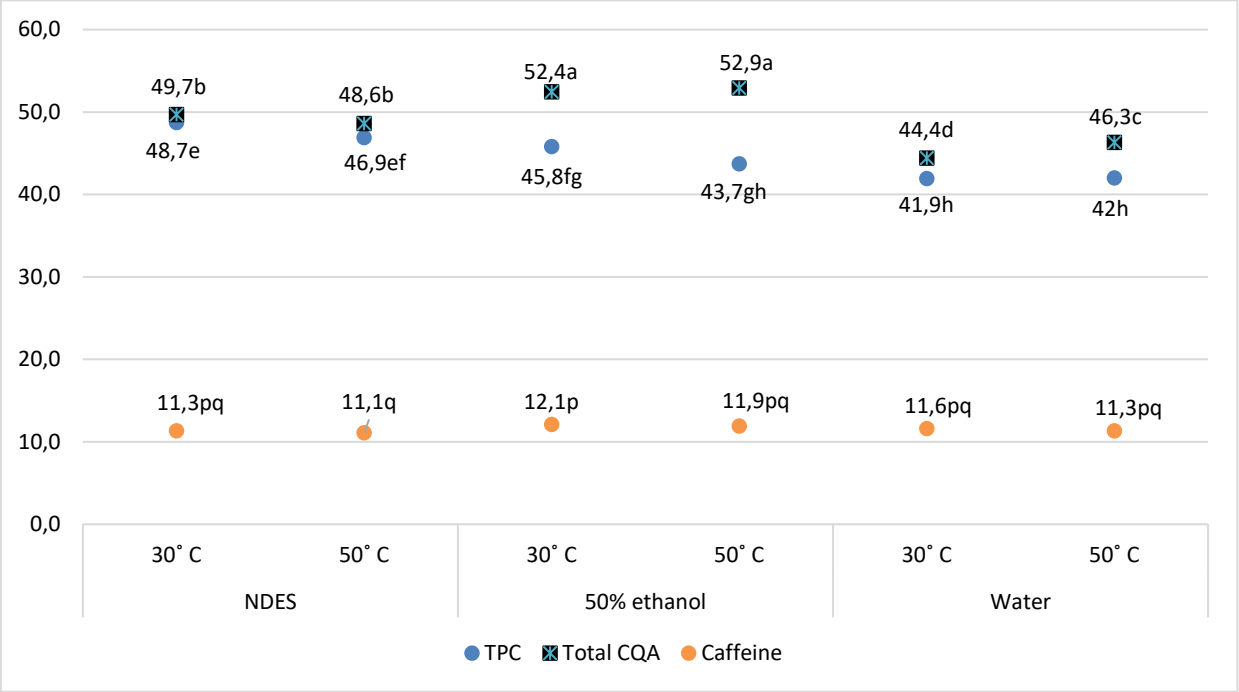

Figure 5: The effect of temperature increase (from 30 °C to 50 °C) on TPC, total CQA and caffeine content of GCB extracts (30 min extraction time)

Values denote the mean of the three replicates based on one-way ANOVA. Means followed by the same letter are not significantly different at a 95 % confidence level according to Tukey's HSD test.

A summary of TPC, chlorogenic acids, and caffeine in glucose-glycerin-based NDES, 50 % ethanol, and water extracts at 30 °C for 30, 60, and 90 min, and at 50 °C for 30 min, is shown in *Table 1*.

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Table 1: Summary of phenolic compounds, caffeine content in NDES, ethanol and water extracts of GCBs

Solvent	Sample Mass (g)	Solvent Volume (mL)	Dilution	Extraction Temperature (°C)	Extraction Time (min)	TPC (mg GAE/g)	5-CQA (mg/g)	3-CQA (mg/g)	4-CQA (mg/g)	Caffeine (mg/g)
NDES (glucose-glycerin)	0.25	5	5	30	30	48.7 ± 0.6^{a}	38.9 ± 0.9^{b}	5.0 ± 0.1^{a}	5.8 ± 0.2^{b}	11.3 ± 0.3^{b}
					60	47.4 ± 0.1^{ab}	39.2 ± 0.6^{ab}	5.0 ± 0.1^{a}	5.9 ± 0.2^{ab}	11.4 ± 0.2^{ab}
					90	48.6 ± 1.1^{a}	39.8 ± 0.4^{ab}	5.0 ± 0.1^{a}	6.0 ± 0.1^{ab}	11.5 ± 0.2^{ab}
				50	30	46.9 ± 0.5^{ab}	38.1 ± 0.6^{b}	4.8 ± 0.1^{a}	5.7 ± 0.1^{b}	11.1 ± 0.2^{b}
50 % ethanol	0.25	5	5	30	30	45.8 ± 1.1^{b}	41.0 ± 0.5^{a}	5.2 ± 0.1^{a}	6.2 ± 0.1a	12.1 ± 0.2^{a}
					60	43.4 ± 0.8^{cd}	40.1 ± 0.6^{ab}	5.1 ± 0.1^{a}	6.0 ± 0.1^{ab}	11.8 ± 0.2^{ab}
					90	45.0 ± 0.5^{bc}	40.8 ± 0.6^{ab}	5.2 ± 0.1^{a}	6.1 ± 0.1^{a}	11.9 ± 0.2^{ab}
				50	30	43.7 ± 0.5^{cd}	41.5 ± 0.9^{a}	5.2 ± 0.1^{a}	6.2 ± 0.1^{a}	11.9 ± 0.2^{ab}
Water	0.25	5	5	30	30	41.9 ± 1.1^{de}	34.8 ± 1.1^{c}	4.4 ± 0.1^{b}	5.2 ± 0.2^{c}	11.6 ± 0.4^{ab}
					60	43.0 ± 0.8^{de}	34.4 ± 0.6^{c}	4.4 ± 0.1^{b}	5.2 ± 0.1^{c}	11.6 ± 0.2^{ab}
					90	40.9 ± 0.8^{e}	34.3 ± 0.7^{c}	4.4 ± 0.1^{b}	5.2 ± 0.1^{c}	11.5 ± 0.2^{ab}
				50	30	42.0 ± 1.3^{de}	36.2 ± 0.2^{bc}	4.6 ± 0.1^{ab}	5.5 ± 1.0^{c}	11.3 ± 0.3^{b}

Values denote the mean ± SD of the three replicates based on one-way ANOVA. Means followed by the same superscript letter within the same column are not significantly different at a 95 % confidence level according to Tukey's HSD test.

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4. DISCUSSION

The solubility of phenolic compounds mainly depends on their chemical structure. For example, the balance between the molecule's polar hydroxyl groups and non-polar aromatic ring determines its solubility. Chemical modifications, such as esterification or glycosylation, can alter the solubility of phenolics (Taheri & Jafari, 2019). Additionally, factors such as temperature, extraction time, pH, and the presence of salts or cosolutes affect their solubility (Furia et al., 2021; Chandimala et al., 2025). Previous studies using COSMO-SAC modelling have shown that phenolic compounds, such as chlorogenic acids, have limited regions available for hydrogen bonding. Likewise, various NDES mixtures exhibit more hydrogen-bonding regions than alcohol (Hijo et al., 2022). The higher TPC yields observed in NDES mixtures compared to those in other solvents may be due to the greater availability of hydrogen-bonding regions in NDES than in water or ethanol. Studies also show that NDES can extract more phenolic compounds due to their higher hydrogen-bonding capacity and improved solute–solvent electrostatic interactions compared to water or pure alcohols (Hijo et al., 2022). Sik et al. (2024) also found that glucose-glycerin-based NDES is more effective than conventional organic solvents (80 % (v/v) ethanol) in extracting phenolic compounds from cornelian cherry. The current study indicated that the highest TPC yield is obtained at 30 min of extraction time. Additionally, TPC yield was lower at 50 °C. These results suggest that a short extraction time at room temperature (~30 °C) is the most suitable for isolating phenolics from GCBs using glucose-glycerin-based NDES.

However, the chlorogenic acid yields in glucose-glycerin-based NDES were comparatively lower than those of ethanol extracts. This can be due to the specific NDES composition used in this study. As a polar molecule, chlorogenic acid shows greater affinity for 50 % aqueous ethanol, which is more polar than the glucose-glycerin mixture. The observed higher caffeine yields in ethanolic extracts may also be due to the higher polarity of 50 % ethanol compared to the specific NDES composition used in this study. In preparing NDES solvent, the ratio of hydrogen bond donors to hydrogen bond acceptors, as well as the addition of water, directly influence the system's polarity and hydrogen bonding capacity, thereby affecting extraction efficiency. Adding water not only reduces viscosity but also improves mass transfer properties and increases polarity, thereby enhancing phenolic yield (Hijo et al., 2022; Loukri et al., 2022). Increasing the solvent volume can also improve plant matrix penetration, but further increases become inefficient. Identifying the ideal solvent volume and dilution level tailored to the NDES and plant material is essential for enhancing extraction efficiency (Taweekayujan et al., 2023). Overall, optimizing glucose-glycerin concentrations in the NDES mixture is crucial to maximize hydrogen bonding, enhancing polarity and phenolic extraction. In the present study, pre-trials conducted with different volumes of a glucose-glycerin mixture (5, 10, and 20 mL) and dilutions (five-fold and ten-fold) revealed that 5 mL of solvent with five-fold dilution yielded the highest TPC. Moreover, heating and stirring NDES components at 90 °C with 50 % water until a clear solution is obtained was found to be the most suitable method for preparing glucose-glycerin-based NDES. Further experiments with other NDES systems, such as choline chloride-glycerin and glucose-lactic acid/citric acid, should be performed to assess their effectiveness in extracting phenolic compounds, including chlorogenic acids, from GCBs.

Further, in this study, total CQA yields exceeded TPC yields in all extracts. This may be because TPC measurements are not specific to all phenolic compounds and include a broad range of phenolics, which may be degraded during extraction. Additionally, the extraction efficiency of different phenolic compounds may vary with the extraction method and conditions used (Platzer et al., 2021).

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5. CONCLUSION

This study was conducted to evaluate the potential of using glucose-glycerin-based NDES for extracting phenolics from GCBs. It found that a glucose-glycerin mixture (1:1 M) with 50 % water is more effective than 50 % ethanol or pure water for isolating phenolics from GCBs, shaking incubation at 30 °C. GCBs extracted with NDES for 30 min yielded the highest TPC (48.7 ± 0.6 mg GAE/g), which was statistically similar to other NDES extracts. NDES for 30 min yielded 49.7 ± 1.2 mg/g of chlorogenic acids and 11.3 ± 0.3 mg/g of caffeine. Chlorogenic acid contents ranged from 43.9 to 44.4 mg/g, 51.1 to 52.4 mg/g, and 49.7 to 50.8 mg/g, in water, ethanolic and NDES extracts, respectively. In NDES for 30 min extract, increasing the extraction temperature to 50 °C resulted in comparatively lower yields of TPC (46.9 ± 0.5 mg GAE/g), *5*-CQA (38.1 ± 0.6 mg/g), *3*-CQA (4.8 ± 0.1 mg/g), *4*-CQA (5.7 ± 0.1 mg/g), and caffeine (11.1 ± 0.2 mg/g). These findings indicate that a short extraction time at room temperature (~30 °C) is most suitable for extracting phenolics from GCBs using glucose-glycerin-based NDES.

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A glükóz-glicerin alapú természetes eutektikus oldószerek alkalmazhatóságának vizsgálata fenolos vegyületek és koffein extrakciójára zöld kávé (*Coffea arabica*) babból

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ÖSSZEFOGLALÁS

Zöld kávébabból (*Coffea arabica*) származó fenolos vegyületek kinyerésére természetes mélyeutektikus oldószer (NDES) keveréket vizsgáltak. A dehidratált, finomra őrölt GCB-ket vízzel, 50 %-os vizes etanollal és NDES-sel (glükóz és glicerin, 1:1 M) extrahálták 30 °C-on 30, 60 és 90 percig rázós inkubációs módszerrel. A folyamatot 50 °C-on 30 percig megismételték. A teljes fenoltartalmat (TPC) Folin-Ciocalteu-teszttel mérték. A klorogénsavak (3-, 4- és 5-koffeoilkinsav) és a koffein mennyiségét HPLC-PDA-val határozták meg. 30 °C-on a legmagasabb TPC-értéket (48,7 ± 0,6 mg GAE/g) a NDES 30 perces kivonatában mérték, ami összehasonlítható ($P > 0,05$) a NDES 60 perces (47,4 ± 0,1 mg GAE/g) és 90 perces (48,6 ± 1,1 mg GAE/g) kivonataival. A legmagasabb klorogénsav-tartalom (52,4 ± 0,6 mg/g) az 50 %-os etanolos, 30 perces extrakcióban volt, ami hasonló ($P > 0,05$) más etanolos kivonatokhoz, valamint a NDES 60 és 90 perces kivonatokhoz. A koffeintartalom (12,1 ± 0,2 mg/g) az 50 %-os etanolos, 30 perces kivonatban érte el a csúcsot, ami hasonló ($P > 0,05$) a legtöbb más kivonathoz, kivéve a NDES 30 perces kivonatot (11,3 ± 0,3 mg/g). Az 50 °C-on végzett NDES 46,9 ± 0,5 mg/g TPC-t, 48,6 ± 0,7 mg/g klorogénsavat és 11,1 ± 0,2 mg/g koffeint eredményezett, ami nem különbözött szignifikánsan a 30 °C-on kapott eredményektől. Ezek az eredmények azt jelzik, hogy a glükóz-glicerin NDES egy ígéretes, környezetbarát oldószer a fenolos vegyületek GCB-kből történő extrakciójához, alternatívát kínálva az 50 %-os etanollal szemben.

Kulcsszavak: klorogénsav, mély eutektikus oldószerek, zöld kávébab, fenolos vegyületek

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